

## IN THE CLAIMS

Please CANCEL claims 1 through 70, without prejudice to the subject matter thereof, and add NEW claims 71-90.

1-70. (Canceled)

71. (New) A recombinant construct, comprising as operably linked components in the 5' to 3' direction, a member selected from the group consisting of:

a seed-specific promoter or a promoter functional in a plant plastid, a DNA sequence encoding a 3-hydroxysteroid oxidase enzyme, and a transcription termination signal sequence;

a seed-specific promoter or a promoter functional in a plant plastid, a DNA sequence encoding a steroid 5 $\alpha$ - reductase enzyme, and a transcription termination signal sequence;

a seed-specific promoter or a promoter functional in a plant plastid, a DNA sequence encoding a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a transcription termination signal sequence;

a seed-specific promoter or a promoter functional in a plant plastid, a DNA sequence encoding a sterol methyl transferase enzyme, and a transcription termination signal sequence;

a seed-specific promoter or a promoter functional in plant plastid, a DNA sequence encoding a sterol acyltransferase enzyme, and a transcription termination signal sequence; and

a seed-specific promoter or a promoter functional in a plant plastid, a DNA sequence encoding an S-adenosylmethionine-dependent  $\gamma$ -tocopherol methyltransferase enzyme, and a transcription termination signal sequence.

72. (New) The recombinant construct of claim 71, further comprising a transit peptide coding region capable of directing transport of said enzyme into a plastid operatively linked to said DNA sequence downstream from said seed-specific promoter.

73. (New) The recombinant construct of claim 71, further comprising:

a gene encoding a selectable marker for selection of plant cells comprising a plastid expressing said selectable marker, and

DNA regions of homology to the genome of said plastid expressing said selectable marker, wherein said DNA regions of homology flank said promoter functional in a plant plastid, said DNA sequence, said transcription termination signal sequence, and said gene encoding a selectable marker downstream from said promoter functional in a plant plastid.

74. (New) The recombinant construct of claim 71, further comprising a ribosome binding site joined to said promoter functional in a plant plastid.

75. (New) The recombinant construct of claim 74, wherein said ribosome binding site is obtainable from a leader sequence selected from a ribosome binding site derived from the group consisting of a plastid leader sequence, a bacterial leader sequence, and a bacteriophage leader sequence.

76. (New) The recombinant construct of claim 74, wherein said ribosome binding site is the ribosome binding site selected from the group consisting of a gene 10 leader sequence and a rbcLRBS ribosome binding site.

77. (New) A plant comprising introduced DNA selected from the group consisting of:

DNA encoding a 3-hydroxysteroid oxidase enzyme, wherein said introduced DNA is operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNA;

DNA encoding a steroid 5 $\alpha$ -reductase enzyme, wherein said introduced DNA is operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and wherein seeds of said plant contain an elevated level of sitostanol, at

least one sitostanol ester, or a mixture thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNA;

DNAs encoding a 3-hydroxysteroid oxidase enzyme and a steroid 5 $\alpha$ -reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme and a tocopherol biosynthetic enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, and at least one tocopherol compound, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a steroid 5 $\alpha$ -reductase enzyme and a tocopherol biosynthetic enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, and at least one tocopherol compound, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a steroid 5 $\alpha$ -reductase enzyme, and a tocopherol biosynthetic enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, and at least one tocopherol compound, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNA encoding a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNA is operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one

phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNA;

DNAs encoding a 3-hydroxysteroid oxidase enzyme and a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a steroid 5 $\alpha$ -reductase enzyme and a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a steroid 5 $\alpha$ -reductase enzyme, and a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a sterol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as a reduced level of campesterol, a

campesterol ester, campestanol, a campestanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a steroid 5 $\alpha$ -reductase enzyme, a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a sterol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as a reduced level of campesterol, a campesterol ester, campestanol, a campestanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs; and

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a steroid 5 $\alpha$ -reductase enzyme, a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a sterol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as a reduced level of campesterol, a campesterol ester, campestanol, a campestanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs.

78. (New) The plant of claim 77, wherein a seed of said plant comprises at least one of the group of brassicastanol, brassicastanol ester, stigmastanol and stigmastanol ester.

79. (New) The plant of claim 77, wherein said regulatory signals cause seed-specific expression of said introduced DNAs, and wherein each of said introduced DNAs is further operatively linked to a transit peptide coding region capable of directing transport of said enzyme encoded thereby into a plastid.

80. (New) The plant of claim 77, wherein said regulatory signals cause plastid-specific expression of said introduced DNAs.

81. (New) The plant of claim 77, further comprising introduced DNA encoding a sterol acyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein a seed of said plant contains an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs.

82. (New) The plant of claim 77, further comprising introduced DNA encoding an S-adenosylmethionine-dependent  $\gamma$ -tocopherol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as an elevated level of  $\alpha$ -tocopherol, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs.

83. (New) The plant of claim 77, that produces a seed having an elevated level of a compound selected from the group consisting of sitosterol, at least one sitosterol ester, sitostanol, at least one sitostanol ester, and mixtures thereof, as well as a reduced level of a compound selected from the group consisting of campesterol, a campesterol ester, brassicasterol, a brassicasterol ester, campestanol, a campestanol ester, brassicastanol, brassicastanol ester, and mixtures thereof, relative to a corresponding transgenic or non-transgenic plant that does not contain introduced DNA encoding a peptide, polypeptide, or protein that affects phytosterol or phytostanol biosynthesis and accumulation in said corresponding plant.

84. (New) The plant of claim 77, wherein said seed contains an elevated level of  $\alpha$ -tocopherol.

85. (New) The plant of claim 77, wherein said at least one phytostanol is sitostanol, and said at least one phytostanol ester is a sitostanol ester.

86. (New) Oil containing a compound selected from the group consisting of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, and mixtures thereof, extracted from seeds of a plant.

87. (New) The oil of claim 86, wherein said oil is produced by a plant comprising introduced DNA selected from the group consisting of:

DNA encoding a 3-hydroxysteroid oxidase enzyme, wherein said introduced DNA is operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNA;

DNA encoding a steroid 5 $\alpha$ -reductase enzyme, wherein said introduced DNA is operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNA;

DNAs encoding a 3-hydroxysteroid oxidase enzyme and a steroid 5 $\alpha$ -reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme and a tocopherol biosynthetic enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, and

at least one tocopherol compound, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a steroid 5 $\alpha$ -reductase enzyme and a tocopherol biosynthetic enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, and at least one tocopherol compound, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a steroid 5 $\alpha$ -reductase enzyme, and a tocopherol biosynthetic enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of sitostanol, at least one sitostanol ester, or a mixture thereof, and at least one tocopherol compound, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNA encoding a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNA is operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNA;

DNAs encoding a 3-hydroxysteroid oxidase enzyme and a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a steroid 5 $\alpha$ -reductase enzyme and a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein



seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a steroid 5 $\alpha$ -reductase enzyme, and a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a sterol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as a reduced level of campesterol, a campesterol ester, campestanol, a campestanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs;

DNAs encoding a steroid 5 $\alpha$ -reductase enzyme, a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a sterol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as a reduced level of campesterol, a campesterol ester, campestanol, a campestanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs; and

DNAs encoding a 3-hydroxysteroid oxidase enzyme, a steroid 5 $\alpha$ -reductase enzyme, a 3-hydroxy-3-methylglutaryl-CoA reductase enzyme, and a sterol methyltransferase enzyme, wherein said introduced DNAs are operatively linked to regulatory signals that cause seed-

specific or plastid-specific expression of said introduced DNAs, and wherein seeds of said plant contain an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, as well as a reduced level of campesterol, a campesterol ester, campestanol, a campestanol ester, or mixtures thereof, compared to seeds of an otherwise identical plant, which does not comprise said introduced DNAs.

88. (New) The oil of claim 86, wherein said oil is produced by a plant comprising at least one introduced DNA sequence encoding a peptide, polypeptide, or protein that affects the biosynthesis and accumulation of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof,

wherein said introduced DNA is operably linked to regulatory signals that cause seed-specific or plastid-specific expression of said introduced DNA, and

wherein said plant produces seed having an elevated level of at least one sterol, at least one phytosterol, at least one phytosterol ester, at least one phytostanol, at least one phytostanol ester, or mixtures thereof, compared to a corresponding transgenic or non-transgenic plant that does not contain said introduced DNA.

89. (New) The oil of claim 86, wherein said oil is produced by a plant that produces a seed having an elevated level of a compound selected from the group consisting of sitosterol, at least one sitosterol ester, sitostanol, at least one sitostanol ester, and mixtures thereof, as well as a reduced level of a compound selected from the group consisting of campesterol, a campesterol ester, brassicasterol, a brassicasterol ester, campestanol, a campestanol ester, brassicastanol, brassicastanol ester, and mixtures thereof, compared to a corresponding transgenic or non-transgenic plant that does not contain introduced DNA encoding a peptide, polypeptide, or protein that affects phytosterol or phytostanol biosynthesis and accumulation in said corresponding plant.

90. (New) The oil of claim 86, wherein said oil is produced by a plant that produces a seed having a reduced level of a compound selected from the group consisting of campesterol, a

campesterol ester, brassicasterol, a brassicasterol ester, campestanol, a campestanol ester, brassicastanol, a brassicastanol ester, and mixtures thereof, compared to a corresponding transgenic or non-transgenic plant that does not contain introduced DNA encoding a peptide, polypeptide, or protein that affects phytosterol or phytostanol biosynthesis and accumulation in said corresponding plant.